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(54) Title: METHOD FOR COMBINATORIAL MATERIALS DEVELOPMENT

(57) Abstract

The present invention provides methods for combinatorial materials development comprising the steps of: (1) creating a set of initial points, (2) testing the set of points according to a determined definition of fitness, (3) choosing a subset of the points based on the selection criteria, (4) perturbing points in the subset until a new larger set is generated that satisfies any determined constraints, and (5) repeating steps 2 and forward until an acceptable set of points is found.

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METHOD FOR COMBINATORIAL MATERIALS DEVELOPMENT

FIELD OF THE INVENTION

The present invention is in the field of biological and chemical synthesis and processing. The present invention relates to methods for developing materials featuring the combination of more than one components. The present invention may be applied in the field of, but is not limited to, the field of chemical or biological synthesis, diagnostics and therapeutics.

The present application claims priority to U.S. Provisional Application Serial No. 60/116,955 filed January 25, 1999.

BACKGROUND OF THE INVENTION

There are many situations where it is desirable to generate a large number of compounds to test for efficacy. Such compounds may be composed of one or more combined materials, *e.g.*, nucleotides, amino acids, chemical moieties, etc. Time constraints are a tremendous obstacle to such an endeavor. Examples of such situations include the generation of polypeptides as drug candidates, the generation of small molecules as drug candidates, and the generation of electrode materials for use in batteries. For example, in the process for discovery of drug candidates, it is normal to produce sets of different polypeptides and then determine how well these polypeptides bind to a receptor of interest. Once a polypeptide is found from among the test set or sets that binds well to a receptor, such polypeptide is pursued as a drug candidate or a drug-precursor candidate.

The number of polypeptides that may be made is enormous thereby making it problematic to test them all, even if the testing is done in a massively parallel way. For instance, if 10-mer polypeptide sequences are made from a set of 20 amino acids, there are 20^{10} (*i.e.*, about 10^{13}) different peptide chains that can be made. Even if a million polypeptides are tested at a time, and each set of a million takes only a minute to test, the total time to test all the polypeptides for desired binding is about 19 years at 24 hours per day, 365 days per year.

Hence, it can take an inordinate amount of time to search every possible polypeptide. It is an object of the present invention to expedite this process. Optimizing can help focus on a region of interest thereby necessitating testing only a small subset of all possible polypeptides. Similar principles of optimizing may be applied to other combinatorial materials thereby providing an efficient and economical means for developing materials comprising one or more parts in combination.

SUMMARY OF THE INVENTION

The present invention provides methods for developing combinatorial materials comprising the steps of:

1. Creating a set of initial points;
2. Testing the set of points according to a determined definition of fitness;
3. Choosing a subset of the points based on the selection criteria;
4. Perturbing points in the subset until a new larger set is generated that satisfies any determined constraints; and
5. Repeating steps 2 and forward using this new larger set.

DETAILED DESCRIPTION OF THE INVENTION

The present invention provides methods for combinatorial materials development. The methods are applicable generally in situations where it is practical to generate only a small portion of the total available materials for testing of efficacy. The methods of according to the present invention comprise the steps of:

1. Creating a set of initial points;
2. Testing the set of points according to a determined definition of fitness;
3. Choosing a subset of the points based on the selection criteria;
4. Perturbing points in the subset until a new larger set of points is generated that satisfies any determined constraints; and
5. Repeating steps 2 and forward using this new larger set.

In preferred embodiments, it is useful to employ the concept of scale of the perturbation as described in the definitions listed below. In some instances, it is possible that the initial set is chosen without clear insight of the best points. Thus, it is often useful to begin with a widely dispersed set of points in the space. As the method according to the present invention proceeds, it selects regions of the space having better fitness than other regions. If the scale of perturbation is then reduced, perturbation results in points closer to each other. For example, if the scale of perturbation is reduced as the process proceeds, the process narrows in on regions of interest causing the process to explore these regions in a more detailed manner, eliminating larger perturbations that would create points outside the regions of interest that would then be rejected in the next ranking of points based on fitness. It is also possible that, if the process enters in a region that is not particularly good according to its measure of fitness, the scale of perturbation can be increased in order to jar the process out of this less-favorable region. For such reasons, it is sometimes useful to adjust the scale of the perturbation as the method according to the present

invention proceeds. Hence, a preferred method according the present invention comprises the steps of:

1. Creating a set of initial points;
2. Choosing a scale of perturbation;
3. Testing the set of points according to a determined definition of fitness;
4. Choosing a subset of the points based on the selection criteria;
5. Perturbing the points in the subset thereby generating a new larger set of points satisfying determined criteria; and
6. Repeating steps 2 and forward until an acceptable set of points is found.

The methods according to the present invention are especially applicable in situations where it is desirable to prepare a set of polypeptides and test for binding of such polypeptides. For instance, according to the methods of the present invention, it is possible to test a first set of polypeptides for binding affinity to a receptor. Of this first set tested, it is possible to screen those polypeptides having the highest binding affinity. Next, it is possible to perturb these highest-affinity polypeptides thereby creating a second test set of polypeptides. Next, it is possible to screen the second set of polypeptides, perturb the resulting highest-affinity polypeptides to create a third test set of polypeptides until one or more polypeptides having a binding affinity that is equal to or better than desired is located. Following such a method may provide polypeptides having optimal binding affinities in a rapid and efficient manner.

The methods according to the present invention are also applicable in situations where it is desirable to obtain small molecules having a desired effect or property. The total number of all possible small molecules is enormous, and in most instances it is not feasible to test all possible small molecules for efficacy as a drug candidate. According to the methods of the present invention, it is possible to provide a first set of small molecules, test the efficacy of the first set of small molecules according to a defined criteria. Next, according to the methods of the present invention it is possible to generate a second test set of small molecules by perturbing a portion of the first set of small molecules demonstrating optimal efficacy according to the first defined criteria.

The methods according to the present invention are also applicable in situations where it is desirable to optimize electrode-materials, e.g. compounds and alloys. Here examples of perturbation include changing the ratio of compounds in an alloy, substituting one or more new compounds, removing a compound, layering compounds, etc.

In another aspect, the present invention features a programmed computer system for implementing a method for developing combinatorial materials. Such a programmed computer system may comprise any one of a large number of possible software programs that may be designed by those skilled in the art. Such a programmed computer system comprises a means for: (1) determining a set of initial points; (2) optionally choosing a scale of perturbation; (3) testing the set of points according to a determined definition of fitness or receiving from some other equipment or entered by hand a list of fitness values associated with the points; (4) choosing a subset of the points based on the selection criteria; (5) perturbing the points in the subset thereby generating a new larger set of points satisfying determined criteria; and (6) repeating the whole process from steps (2) on until an acceptable set of points is found.

As used herein, the following terms are understood to mean the following:

A "space of interest" is a set of all possible particular samples that may be examined or tested according to one or more criteria. For example, in the situation where it is desirable to isolate polypeptides having a particular property, the space of interest is all polypeptides that can be made. In the situation where it is desirable to isolate battery electrode alloys having a particular property, the space of interest is all alloys that can be made. In the situation where f one is interested in a parameter as represented by a real number between 0 and 10, the space of interest is all real numbers between 0 and 10.

A "point" in a space of interest is a particular sample in the space of interest. For example, one particular polypeptide is a point in the space of interest comprising all polypeptides. One particular alloy is a point in the space of interest comprising all alloys. A particular number, for example 6, is a point in the space of interest comprising all numbers from 0 to 10.

"Perturbation" is the process of changing one point from a space of interest thereby creating a new point in the space of interest. For example, it is possible to perturb the number 6.18 by adding 0.01 thereby creating 6.19, a new point in the space of interest comprising real numbers from 0 to 10. A DNA strand defined by GATTACA may be perturbed by changing its third position from a T to an A thereby creating a new DNA strand defined by GAATACA. Likewise, a DNA strand may be perturbed by adding additional nucleotides or removing nucleotides thereby producing different DNA sequences. Perturbing of an alloy may feature increasing the fraction of one metal or adding a new metal into the mix. Similarly, perturbing a polypeptide may feature adding additional amino acids to the sequence, removing particular

amino acids from the sequence or substituting another amino acid for one or more amino acids in the sequence.

Perturbation may be limited by "constraints" on the space of interest. For example, polypeptides of 10 amino acids or less in length may be of interest. In such a situation, it is not desirable to explore the space outside these constraints. Hence, it is not desirable to choose any perturbations that create a point outside the constraints (such as a polypeptide of length 11). An exemplary constraint on alloys might be that the alloy should not contain both aluminum and zinc at the same time. An exemplary constraint on DNA space might be that the DNA strands are longer than 10 nucleotides and shorter than 50 nucleotide bases.

The space of interest may have a "metric" or "distance function" such that any two points in the space have a corresponding scalar that is used to provide a distance between the two points. For example, two points on a road map have a distance between them – the "metric" can be a measure of distance in meters, for example. For polypeptides, the metric may be the minimum number of peptide positions that do not match up.. For alloys, the metric may be measured by $\text{sum}_k(a_k - b_k)^2$, where sum_k represents a sum over k, k is an index that runs over all metals present in the alloys, a_k is the fraction of metal k present in the first alloy and b_k is the fraction of metal k present in the second alloy. For a set of real numbers, a metric might be $(n1 - n2)^2$, where n1 is the first number and n2 is the second number.

In situations where the space has a metric, the perturbation may have a "scale" such that if a perturbation with one scale results in a new point in the space a distance D from the original point (as measured by the metric), a perturbation of the same point with a larger scale on average results in a new point in the space a distance greater than D from the original point. A particular perturbation may or may not be substantially random.

"Fitness" generally means how good or bad a particular point is in accordance with the defined criteria. For example, regarding a polypeptide, "fitness" may describe how strongly the polypeptide binds its target. In the case of real numbers, "fitness" may describe how close a particular function of that number is to a target value. In the case of metal alloys, "fitness" may describe how well the current/voltage curve of the alloy matches some desired current/voltage curve or how well it scores on a rating of its current/voltage curve.

"Selection criteria" are used to select a subset of points based on how they are ranked by fitness. Exemplary "selection criteria" include selecting N best points as ranked by fitness. Further exemplary "selection criteria" may include eliminating the best M points and choosing

then next N best ones. Exemplary "selection criteria" may include picking randomly but with a higher probability of picking optimal points. For example, where $y = (f - f_{min}) / (f_{max} - f_{min})$, where f_{min} is the minimum fitness number, f_{max} is the maximum fitness number, and f is the fitness number of the point being considered, a number between 0 and 1 may be generated randomly and determined whether to be lower than y. If so, that point may be chosen. If not, the next point for selection is considered. This process, for example, may be continued until N points are chosen from the original set.

DESCRIPTION OF THE PREFERRED EMBODIMENTS

The following are provided purely by way of example and are not intended to limit the scope of the present invention.

EXAMPLE 1

Arrays that synthesize chemicals including arrays that synthesize DNA are being developed to synthesize and process information. Chemical synthesis and processing on arrays of electrodes depends upon various parameters such as buffer concentration, percent acetonitrile in the buffer, length of time for deblocking chemistry, and the total voltage-on time to use for a square-wave voltage signal during deblocking.

Where genetic material such as DNA is being synthesized, the goal of the synthesis is to produce high quality DNA. Determining how to optimize the various parameters to produce optimal DNA synthesis may require much effort. The methods according to the present invention were employed to determine optimal conditions.

Our space here is the possible set of parameters -- each one being a real number between 0 and infinity. A point in the space is a particular set of parameters, such as setting the buffer concentration to 0.0083 molar, setting the ACN percentage at 25%, running the deblocking for 827 seconds, and setting voltage-on time to 223 seconds. This point can be represented by (ln(0.0083), 25, 827, 223). We chose to use the natural log of the concentration as that better represents that we are evaluating an order of magnitude on the number.

We chose to perturb points in this space by, for each parameter, selecting a number at random from a gaussian distribution centered at zero and with a standard deviation equal to half a rough estimate of the maximum useful value minus the minimum useful value. For deblock, sigma was 500 seconds; for concentration, sigma was 1.5; for percentage, sigma was 50; for voltage on, sigma was 500 seconds.

The constraints on perturbation were that we didn't want values less than one number and larger than another. We did not accept concentration numbers less than 0 or greater than 1 molar;

ACN percentage less than 0 or greater than 100; deblock time less than 30 seconds or greater than 3000 seconds; and voltage-on time less than one quarter the deblock time or greater than the deblock time.

The metric picked was the euclidean distance between points, *i.e.*, sqrt((v1-w1)^2 + (v2-w2)^2 + ... + (vN-wN)^2), where vk and wk are the kth component of the vector. Thus the distance between (ln(0.0083), 25, 827, 223) and (ln(0.1), 50, 400, 120) is sqrt((-4.791+2.303)^2 + (25-50)^2 + (827-400)^2 + (223-120)^2).

The scale of perturbation was defined by the sigmas of the gaussians used in perturbing and just remained fixed throughout the process.

The fitness was determined by the quality and brightness of florescent spots generated by the oligonucleotides built with florescent tags in the bases. Humans were the judges of the quality.

The selection criterion was to take the best point (as ranked by fitness) and to keep perturbing it until we found a better point. Then we keep perturbing that until we found a better point, and so on.

This resulted rather quickly in finding parameters that worked acceptably well according to our fitness criterion without having to test an enormous grid of possible parameter settings and including the variability of human judgement when coming up with fitness of points.

EXAMPLE 2

Polypeptide Drug Candidates

The space of interest is polypeptides composed of the 20 natural amino acids and from five to ten amino acids in length. This comprises the specification for the space and constraints. These polypeptides may be constructed electrochemically on an electrochemical array chip; chemically on beads, pins, or substrates; made in microfluidics chambers; etc.

The perturbation in this instance may include the following basic operations: (1) addition of an amino acid (adding one at the front of the polypeptide, inserting one somewhere in the middle, or adding one to the end), (2) the deletion of an amino acid, or (3) changing an amino acid.

The metric is how many of these basic operations are applied to get from one polypeptide to another. The scale of perturbation can likewise be how many basic operations were used in the perturbation.

Fitness is chosen to be the binding coefficient determined as follows. For the reaction A + B <-> AB, where A is the receptor of interest, B is the polypeptide candidate, and AB is the

polypeptide bound to the receptor, a binding coefficient is defined as K = [AB]/([A][B]), where [x] is the concentration of x. A spot containing polypeptide B may be exposed to a solution containing receptor A, which is florescently labeled, thereby providing an indication of how strongly A binds to B by the brightness of the resulting spot. So, [AB] can be estimated as being proportional to the intensity of the spot; [B] may be varied by how much B is built at a spot; and [A] may be kept constant. Then, assuming K is a constant, it is possible to estimate its size for one polypeptide compared to another by looking at K = d([AB])/d[B] x (1/[A]), such as by plotting [AB] vs. [B], taking the slope and dividing by [A]. We can do this by building a dilution series of B on a synthesis array, for example, building several spots of the same polypeptide B at different concentrations on the surface. This dilution-series method helps avoid false positives and can allow picking a fitness function such that one is looking for a particular size of K -- neither too high nor too low -- a fitness function equal to $(K - Ktarget)^2$. There are other fitness functions that may be chosen. It is possible to choose how well the polypeptide binds to the receptor of interest as judged optically by having the receptor florescently labeled and examining which spots on an array of polypeptides light up the strongest after applying a solution containing the receptor to the polypeptide array. It is possible to avoid false positives and false negatives to some extent by building the same polypeptide at several different sites and eliminating the darkest and the brightest then taking the mean of the rest to come up with a fitness number.

Set the selection criterion to be that one takes the best 10% of the polypeptides generated.

Choose to keep the selected points in the new set, filling out the set with perturbed versions where the perturbation is chosen at random from the set of the three basic operations but applying only one or two basic operations to generate a perturbation thereby limiting the scale of the perturbation.

Thus, an initial set of polypeptides would be generated, perhaps as perturbed versions of some currently known polypeptide of some functionality or perhaps creating a set at random from the space of interest. It is possible to create four spots on an array for each of the polypeptides being tested. Then it is possible to apply the labeled receptor, measure the brightness of the spots, eliminate the high and low for each spot, and take the mean of the rest thereby providing fitness numbers for each polypeptide. It is then possible to take the best 10%, perturb to fill out a new array, test, and so on.

EXAMPLE 3

Small-Molecule Drug Candidates

The space of interest is small molecules. Such small molecules may be generated by an electrochemical array; synthesized on beads, pins, or some other substrate; or synthesized in microfluidics chambers; etc.

Perturbation may be one of the following basic operations: adding a chemical compound to the molecule's scaffold, deleting a chemical compound from the molecule's scaffold, changing a molecule on the scaffold, adding another scaffold to the existing scaffold, changing the chirality of the scaffold, creating a loop from one terminus of the scaffold to another, or inserting a metal into the scaffold.

The metric may be how many of these basic operations are applied to get from one small molecule to another. Another possible metric is a similarity measure that measures molecular diversity.

The scale of perturbation may be how many basic operations were used in the perturbation.

Fitness may be how well the small molecule binds to the protein of interest as judged optically by having the protein florescently labeled and determining which spots on an array of small molecules light up the strongest after applying a solution containing the protein to the small molecule array. One may avoid false positives and false negatives by building the same small molecules at several different sites and removing the darkest and the brightest then taking the mean of the rest to come up with a fitness number. One could also have chosen to use the binding coefficient fitness function (based on a dilution series) as described in the "polypeptide drug candidates" example.

The selection criterion is the best 10% of the small-molecules generated.

The selected points are maintained in the new set, filling out the set with perturbed versions where the perturbation is chosen at random from the set of the basic operations but applying only three or less basic operations to generate a perturbation thereby limiting the scale of the perturbation.

It is possible to generate an initial set of small molecules, perhaps as perturbed versions of some currently known small molecule of some functionality or perhaps creating a set at random from the space of interest. It is possible to create four spots on an array for each of the small molecules being tested. Then it is possible to apply the labeled protein, measure the brightness of the spots, throw out the high and low for each spot, and take the mean of the rest. This generates fitness numbers for each small molecule. It is then possible to takes the best 10%, perturbs to fill out a new array, tests, and so on.

EXAMPLE 4

Chelating Material

Molecules that form chelates with metal ions are often characterized by their absolute and relative affinity for different metal ions. Some uses require chelating molecules that will bind to all metal ions in a non-specific manner. Other uses require a chelating molecule that has a very specific affinity for a particular metal ion and not for other metal ions.

Chelating molecules can be formed from many different organic, inorganic and mixed organic/inorganic motifs. For example, chelating molecules can be formed from polyethers, from pyridine oligomers, or from small peptides. Other materials include cyclic molecules such as ethylene diamine tetra acetate (EDTA) and porphyrin moieties.

Numerous methods can be used to create combinatorial libraries of chelator materials. For example, in instances where the chelator molecule is a small peptide, the peptide may be synthesized on beads, pin arrays, using ink-jet deposition, or on electrode arrays. Similar methods may, for example, be used to modify a porphyrin scaffold molecules to create combinatorial libraries.

The fitness of a chelator molecule can be determined by evaluating its binding selectivity and affinity. This can be accomplished in a variety of ways including testing sample solutions that have been exposed to the chelator molecule for unbound metal ions. For example, beads from a combinatorial chelator library may be added to individual wells in a microtiter plate and exposed to a solution containing the metal ion of interest as well as other interfering ions. The solutions in each individual well may then be tested for the metal ion or ions of interest using, for example, a colorometric assay. As another example, small peptides made on an electrode array may be exposed to a solution containing one or more metal ions of interest. The quantity and identity of the metal ions that are incorporated into each peptide chelator molecule may be determined electrochemically. Further, binding affinity coefficients may be determined on an electrode array by constructing a dilution series in the candidate peptide chelator molecules by making different amounts at different electrodes in the electrode array. The relative quantity of metal ion or ions sequestered by the candidate chelator may be evaluated electrochemically at each point in the dilution series, which produces a dilution series curve.

After evaluating chelator molecule candidates for selectivity and binding affinity, the top 10% of the candidate materials from a combinatorial chelator molecule library are selected for optimization using perturbation by the methods described previously. Perturbations may be introduced by many methods. For example, different amino acids may be inserted into a peptide chelator sequence or new molecular motifs may be added to a porphyin scaffold moiety. Also,

perturbations may be introduced into the composition of the solution containing the metal ion or ions to which the chelator candidate molecules is exposed. Such perturbations include changes in the solvent composition, spectator electrolytes, interfering ions, pH and other such changes as will be evident to one skilled in the art. These perturbations can help optimize the operating parameters for applications that use the candidate chelator molecules.

EXAMPLE 5

Electrodeposition

Deposition of material by electrochemical methods depends on numerous experimental variables. These variables reflect the complex interplay between such factors as the composition of a plating solution, the electrical parameters and the substrate material or materials. There are also many different desired results from the process of electrodepostion. A metal like platinum may be electrodeposited on a decorative item for aesthetic reasons or may be electrodeposited to form a catalyst.

The search space for optimizing the conditions for electrochemical deposition includes such diverse variables as the composition of electrolytes or additives in a plating solution, the schedule on which electrical variables are applied to the conductive substrate onto which materials are electrodeposited and the nature of the deposition substrate. Combinatorial exploration of this electrodeposition variable space can be accomplished by many means including electrode arrays, mechanical deposition on a conductive substrate using ink-jet or pen nib spotting techniques, photolithographic masking, the use of chemical redox agents among others.

The test metrics of merit depend on the ultimate use of the electrodeposited material. For example, a catalyst material may be evaluated based on its longevity or its catalytic activity, a decorative coating may be evaluated on the basis of its smoothness and luster, a superconducting material on the basis of its superconducting transition temperature.

The top 10% of the candidate electrodeposition protocols from a combinatorial protocol library may be selected for optimization using perturbation by the methods described previously. Perturbations may be introduced by many methods. For example, the voltage applied for electrodeposition may be changed and the length of time that the voltage is applied may be varied. The current or voltage as a function of time may take any form, and these functions may be perturbed in a wide variety of ways. A sinusoidal current signal, for example, may be perturbed by changing its frequency, by adding in additional fourier components, or by introducing or perturbing an amplitude envelop among others. The composition of the solution

from which materials are electroplated may by varied. For examples, the electrolyte composition and concentration may be changed, surfactant additives may be removed or added, the solution may have two or more immisible phases among others.

EXAMPLE 6

Fuel Cell Catalyst

Catalysts for fuel cells are often made of one or more metals. One of these metals is usually a transition metal, such as platinum, rhodium, or ruthenium. There are many different important variables that determine fuel cell catalyst performance. These include alloy composition, surface morphology, alloy phase segregation, crystallinity, doping inclusions and the like. Alloy composition, morphology and other factors derive from a complex interplay between the materials that are deposited in a combinatorial manner and the methods used for their deposition.

Numerous methods may be used to create combinatorial libraries of fuel cell catalyst materials. These methods include physical deposition from ink-jet print heads, mechanical spotting onto substrates using pin-nib spotting, electrodepostition onto electrode arrays, and combinations of these different methods. After deposition, combinatorial fuel cell catalyst candidate materials can be further processed by many methods. These post-processing methods include thermal annealing, electrochemical conditioning, doping and combinations of these methods.

The fitness of a fuel cell catalyst may be tested by many methods. For example, the current density that can be achieved at a given voltage in a test solution is one test. Another test is the longevity of this current density under electrochemical load cycling and the susceptibility of the materials to poisoning from contaminants.

The top 10% of the candidate materials from a combinatorial fuel cell library that are tested may be selected for optimization using perturbation by the methods described previously. Perturbations may be introduced by many methods. For example, varying the ratios of alloys in the deposition solutions may yield different alloy compositions, varying the current-voltage characteristics of the electrochemical deposition protocol may result in a host of perturbations ranging from alloy composition to surface morphology, and varying thermal annealing protocols may result in perturbations of morphology, crystallinity and doping levels among other effects.

EXAMPLE 7

Zinc finger protein DNA binding

The zinc finger motif is an important functional element of many proteins that bind to DNA sequences. These motifs are found in numerous transcription factors and steroid receptor complexes. The primary DNA sequence that these proteins bind to is not always known. Any secondary DNA sequences that these proteins may bind to often is not known. For example, it is often desirable to determine the effects of mutations on a proteins sequence selectivity and binding affinity.

Combinatorial libraries of DNA can be used to determine the binding affinity and selectivity for proteins that carry a zinc finger motif. These libraries can be created by many means. Such means include electrode arrays, photolithographic patterning, mechanical deposition by ink-jet or pen-nib spotting among others.

The affinity and selectivity of one or more proteins carrying a zinc finger motif for a particular sequence of DNA may be determined by many methods. For example, an array of electrodes that have different DNA sequences over different electrodes may be used to detect, to monitor and to quantify any proteins labeled with an electrochemically active tag that bind to the DNA sequence at any particular electrode. As a further example, fluorescence may be used in an analogous manner to detect, to monitor and to quantify any proteins labeled with a fluorescent tag that bind to DNA sequences in arrays of DNA sequences that have been prepared by photolithographic methods. Binding affinity may be evaluated in a facile manner by determining the relative amount of protein that binds to different locations in an array that have different amounts of the same DNA sequence. It may also be desirable to, for example, to determine that effect of a particular DNA sequence on the binding affinity and selectivity of a receptor cofactor such as a steroid hormone.

The top 10% of the candidate sequences from a combinatorial DNA sequence may be selected for optimization using perturbation by the methods described previously. Perturbations can be introduced by many methods. For example, the DNA sequence may be varied by addition, subtraction or substitution of a different nucleic acid into the DNA sequence.

EXAMPLE 8

Efficient phosphor materials

Materials that have phosphorescent properties are often composed of one or more inorganic elements and often are sensitive to trace levels of doped inactivator complexes. These materials have many uses such as flat panel plasma displays. The efficiency, lifetime and

stability of the color output of a phosphor material determine its utility for commercial applications.

Numerous methods may be used to create combinatorial libraries of low voltage phosphor materials. These methods include precipitation from solutions, physical deposition from ink-jet print heads or pen-nib spotting, electrodeposition onto electrode arrays and combinations of these and other methods. After deposition, the materials may be processed in many different ways including thermal and electrical processing, doping and combinations of these methods.

The fitness of a particular material as a low voltage phosphor may be evaluated by numerous methods. For example, the intrinsic phosphorescence of an array of materials may be evaluated by illuminating the array with UV light and measuring the luminescent output from each of the materials. Another method for testing low voltage phosphors involves placing an array of candidate materials in a chamber filled with an inert gas such as Xenon, exciting the gas with a voltage source and measuring the luminescent output from each of the materials in the array.

The top 10% of the candidate materials from a combinatorial phosphor library may be selected for optimization using perturbation by the methods described previously. Perturbations can be introduced by many methods. For example, varying the ratios of elements and dopants in the deposition solutions can yield different alloy compositions. Other perturbations include varying the current-voltage characteristics of an electrochemical deposition protocol, varying the thermal annealing schedule and varying the type and amount of doping introduced after the initial deposition.

Although the invention has been described with reference to the presently preferred embodiments, it should be understood that various modifications can be made without departing from the spirit of the invention. Accordingly, the invention is limited only by the following claims.

WHAT IS CLAIMED IS:

1. A method for developing combinatorial materials comprising the steps of:
   (a) Creating a set of initial points;
   (b) Testing the set of points according to a determined definition of fitness for selection criteria;
   (c) Choosing a subset of the points based on the selection criteria;
   (d) Perturbing points in the subset until a new larger set is generated that satisfies any determined constraints; and
   (e) Repeating steps (b) and forward using this new larger set.

2. The method of claim 1 wherein the materials developed are polypeptides.

3. The method of claim 1 wherein the materials developed are oligomers.

4. The method of claim 1 wherein the materials developed are small molecules.

5. The method of claim 1 wherein the materials developed are electrode materials selected from the group consisting of chemical compounds and alloys.

6. The method of claim 1 wherein the materials developed are chelating agents.

7. The method of claim 1 wherein the materials developed are electrodeposition protocols.

8. The method of claim 1 wherein the materials developed are fuel cell catalysts.

9. The method of claim 1 wherein the perturbing comprises removing one or more nucleotides.

10. The method of claim 1 wherein the perturbing comprises adding one or more nucleotides.

11. The method of claim 1 wherein the perturbing comprises substituting one or more nucleotides.

12. The method of claim 1 wherein the perturbing comprises removing one or more amino acids.

13. The method of claim 1 wherein the perturbing comprises adding one or more amino acids.

14. The method of claim 1 wherein the perturbing comprises substituting one or more amino acids.

15. The method of claim 1 wherein the selection criteria is binding affinity to a receptor molecule.

16. A method for combinatorial materials development comprising the steps of:

(a) Creating a set of initial points;

(b) Choosing a scale of perturbation;

(c) Testing the set of points according to a determined definition of fitness;

(d) Choosing a subset of the points based on the selection criteria;

(e) Perturbing the points in the subset thereby generating a new larger set of points satisfying determined criteria; and

(f) Repeating steps (b) and forward until an acceptable set of points is found.

17. The method of claim 16 wherein the materials developed are polypeptides.

18. The method of claim 16 wherein the materials developed are oligomers.

19. The method of claim 16 wherein the materials developed are small molecules.

20. The method of claim 16 wherein the materials developed are electrode materials selected from the group consisting of chemical compounds and alloys.

21. The method of claim 16 wherein the materials developed are chelating materials.

22. The method of claim 16 wherein the materials developed are electrodeposition protocols.

23. The method of claim 16 wherein the materials developed are fuel cell catalysts.

24. The method of claim 16 wherein the perturbing comprises removing one or more nucleotides.

25. The method of claim 16 wherein the perturbing comprises adding one or more nucleotides.

26. The method of claim 16 wherein the perturbing comprises substituting one or more nucleotides.

27. The method of claim 16 wherein the perturbing comprises removing one or more amino acids.

28. The method of claim 16 wherein the perturbing comprises adding one or more amino acids.

29. The method of claim 16 wherein the perturbing comprises substituting one or more amino acids.

30. The method of claim 16 wherein the selection criteria is binding affinity to a receptor molecule.

31. A combinatorial material produced by the process according to claim 1.

32. A combinatorial material produced by the process according to claim 17.